(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
12 September 2002 (12.09.2002)

PCT

(10) International Publication Number
WO 02/070664 A3

(51) International Patent Classification[7]: C12Q 1/68, 1/70, C12P 19/34, G01N 33/53, 33/566, C07H 21/02, C07K 5/00

(21) International Application Number: PCT/US02/06763

(22) International Filing Date: 4 March 2002 (04.03.2002)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
09/798,007 2 March 2001 (02.03.2001) US

(71) Applicant *(for all designated States except US)*: **ISIS PHARMACEUTICALS, INC.** [US/US]; 2292 Faraday Avenue, Carlsbad, CA 92009 (US).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **ECKER, David, J.** [US/US]; 1041 Saxony Road, Encinitas, CA 92024 (US). **GRIFFEY, Richard, H.** [US/US]; 360 Barsby Street, Vista, CA 92084 (US). **SAMPATH, Rangarajan** [IN/US]; 12223 Mannix Road, San Diego, CA 92129 (US). **HOFSTADLER, Steven** [US/US]; 5014 Viewridge Way, Oceanside, CA 92056 (US). **MCNEIL, John** [US/US]; 427 Retaheim Way, La Jolla, CA 92037 (US).

(74) Agents: **CALDWELL, John, W.** et al.; Woodcock Washburn LLP, One Liberty Place - 46th Floor, Philadelphia, PA 19103 (US).

(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZM, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *with international search report*
— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

(88) Date of publication of the international search report:
19 December 2002

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: METHOD FOR RAPID DETECTION AND IDENTIFICATION OF BIOAGENTS

(57) Abstract: Method for detecting and identifying unknown bioagents, including bacteria, viruses and the like, by a combination of nucleic acid amplification and molecular weight determination using primers which hybridize to conserved sequence regions of nucleic acids derived from a bioagent and which bracket variable sequence regions that uniquely identify the bioagent. The result is a "base composition signature" (BCS) which is then matched against a database of base composition signatures, by which the bioagent is identified.

WO 02/070664 A3

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US02/06763

A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C12Q 1/68, 1/70; C12P 19/34; G01N 33/53, 33/566; C07H 21/02; C07K 5/00

US CL : 435/4, 5, 6, 7.1, 91.1, 91.2; 536/23.1, 24.3; 530/350, 387.1, 388.1; 436/500, 501

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 435/4, 5, 6, 7.1, 91.1, 91.2; 536/23.1, 24.3; 530/350, 387.1, 388.1; 436/500, 501

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Please See Continuation Sheet

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X --- Y	HURST et al. Detection of bacterial DNA polymerase chain reaction products by matrix assisted laser desorption/ionization mass spectrometry. Rapid Communications in Mass Spectrometry. 1996, Vol. 10, pages 377-382, see entire document.	1-3, 5, 6, 8-14, 20-22, 24, 25, 27-34 --- 4, 7, 15-19, 23, 26, 35-48
Y	WELHAM et al. The characterization of microorganisms by Matrix Assisted Laser Desorption/ionization time of flight Mass Spectrometry. Rapid Communications in Mass Spectrometry. 1998, Vol. 12, pages 176-180, see entire document.	1-48
Y	CHO et al. Application of the ribonuclease P (RNAse P) RNA gene sequence for phylogenetic analysis of the genus saccharomonospora. International J. Systematic Biology. 1998, Vol. 48, pages 1223-1230, see entire document.	7, 26
Y	WO 99/31278 A1 (SEQUENOM INC.) 24 June 1999, see entire document.	15-19, 35-38

☒ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
12 August 2002 (12.08.2002)	23 OCT 2002
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703)305-3230	Authorized officer Jeffrey Fredman Telephone No. 703-308-0196

Form PCT/ISA/210 (second sheet) (July 1998)

PCT/US02/06763

INTERNATIONAL SEARCH REPORT

C. (Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	MATRAY et al. Synthesis and properties of RNA analogs - oligoribonucleotides N3' - p5' phosphoramidites. Nucleic Acids Research. 1999, Vol. 27, No. 20, pages 3976-3985, see entire document.	18, 37
Y	U.S. 5,605,798 A (KOSTER) 25 February 1997, see entire document, especially claim 10.	4, 23
X --- Y	LI et al. Single nucleotide polymorphism determination using primer extension and time of flight mass spectrometry. Electrophoresis. 1999, Vol. 20, pages 1258-1265, see entire document.	39, 40, 42, 44-48 ---------- 1-38, 41, 43
X --- Y	WO 98/20166 A2 (SEQUENOM INC.) 14 May 1998, see entire document.	1-6, 8-14, 20-25, 27-34,39-48 ---------- 7, 15-19, 26, 35-38

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US02/06763

Box I Observations where certain claims were found unsearchable (Continuation of Item 1 of first sheet)

This international report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claim Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claim Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. ☐ Claim Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of Item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:
Please See Continuation Sheet

1. ☒ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest.

☒ No protest accompanied the payment of additional search fees.

BOX II. OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING

This International Search Authority has found 2 inventions claimed in the International Application covered by the claims indicated below:

This application contains the following inventions or groups of inventions which are not so linked as to form a single inventive concept under PCT Rule 13.1. In order for all inventions to be searched, the appropriate additional search fees must be paid.

Group I, claim(s)1-38, drawn to methods of detecting unknown biological agents.
Group II, claim(s) 39-48, drawn to methods of detecting single nucleotide polymorphisms.

and it considers that the International Application does not comply with the requirements of unity of invention (Rules 13.1, 13.2 and 13.3) for the reasons indicated below:

The inventions listed as Groups I-II do not relate to a single inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they laek the same or corresponding special technical features for the following reasons: The Groups lack a special technical feature because Hurst et al (Rapid Communications in Spectrometry (1996) 10:377-382) teaches a method of identifying a bioagent comprising: a) contacting nucleic acid from said bioagent with at least one pair of primers (page 378, column 1 and table 1), b) amplifying the nucleic acid sequecne to produce an amplification product (page 378, column 2), c) determiing the molecular mass of the amplification product (see page 378, column 2 and page 379, figure 1), d) comparing the molecular mass to a known organism (page 379, figure 1). This reference anticipates the claim thereby eliminating the special technical feature.

Continuation of B. FIELDS SEARCHED Item 3:

EAST, MEDLINE, BIOSIS, CAPLUS
search terms: bioagent, RNA, DNA, amplify, LCR, SDA, PCR, ligase, polymerase, chain, strand, displacement, molecular, mass, spectrometry, time, flight, quadrupole, analog, diaminopurine, single, nucleotide, polymorphism.